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- (S) Human metabotropic glutamate receptor and related DNA compounds.
- 57 This invention provides a human glutamate receptor and functional equivalents thereof, and nucleic acids compounds which encode the receptor. The invention also provides assays, probes and primers, and other molecular biology techniques which utilize the compounds disclosed.

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This invention relates to a novel human glutamate receptor protein and to novel nucleic acid compounds that encode the novel protein.

In the mammalian central nervous system, L-glutamate serves as a major excitatory neurotransmitter. The interaction of glutamate with its membrane bound receptors is believed to play a role in many important neuronal processes including fast synaptic transmission, synaptic plasticity, and longterm potentiation. These processes are fundamental to the maintenance of life and normal human abilities such as learning and memory. Monaghan, D. T. et al., 8 Neuron 267 (1992).

Pharmacological characterization of receptors for L-glutamate has led to their classification into two families based on their biological function: the ionotropic receptors which are directly coupled to cation channels in the cell membrane, and the metabotropic receptors which function through coupling to G-proteins. The present invention concerns a member of the metabotropic family of glutamate receptors.

In addition to its role in normal human physiology, interaction of L-glutamate with its receptors is believed to play a key role in many neurological disorders such as stroke, epilepsy, and head trauma, as well as neurodegenerative processes such as Alzheimer's disease. Olney, R. W., 17 *Drug Dev. Res.* 299 (1989). For this reason, understanding the molecular structure of human L-glutamate receptors is important for understanding these disease processes as well as for searching for effective therapeutic agents. Up to the present, the search for therapeutic agents which will bind and modulate the function of human glutamate receptors has been hampered by the unavailability of homogeneous sources of receptors. The brain tissues commonly used by pharmacologists are derived from experimental animals (non-human) and furthermore contain mixtures of various types of glutamate receptors.

In searching for drugs for human therapy, it is desirable to use receptors which are more analogous to those in the intact human brain than are the rodent receptors employed to date. The discovery of human glutamate receptors, therefore, provides a necessary research tool for the development of selective pharmaceutical agents. The present invention provides a human glutamate receptor, HSmGluR1, which can be used to search selectively for drugs which modulate this receptor.

Recently, four metabotropic receptor subtypes (mGluR1-mGluR4) have been cloned from rat brain. Masu *et al.*, 349 Nature 760 (1991); Houamed *et al.*, 252 Science 1314 (1991); and Tanabe Y. *et al.* 8 Neuron 169 (1991). In addition, two alternately spliced versions of mGluR1 are known. Tanabe Y. *et al.* 8 Neuron 169 (1991).

The present invention provides compounds which comprise the amino acid sequence SEQ ID NO:1 or a functional equivalent thereof. In particular, the amino acid compound which is SEQ ID NO:1 is preferred.

The invention also provides nucleic acid compounds which comprise a nucleic acid sequence which encodes the amino acid compounds provided. Particularly, nucleic acid compounds which are DNA are preferred. Most preferred is the DNA compound SEQ ID NO:2. However, also preferred are those nucleic acid compounds which are sense mRNA.

Also provided by the present invention are recombinant nucleic acid vectors comprising the nucleic acids which encode SEQ ID NO:1. The preferred nucleic acid vectors are those which are DNA. Most preferred are recombinant DNA vectors which comprise the DNA sequence which is SEQ ID NO:2. A preferred DNA vector which comprises SEQ ID NO:2 is pRS117.

Moreover, recombinant DNA vectors of the present invention preferably comprise a promoter positioned to drive expression of a DNA sequence which encodes SEQ ID NO:1. Those vectors wherein said promoter functions in mammalian cells are preferred. Those mammalian vectors wherein said promoter functions in AV12 cells are preferred. The recombinant DNA expression vector most preferred is plasmid pRS121.

Restriction fragments of the preferred vectors are also provided. Particularly, the approximately 4.1 kb EcoRI and the approximately 3.8 kb BssHII/AfIII restriction fragment of a vector which comprises SEQ ID NO:2 are provided.

The present invention also provides probes and primers useful for molecular biology techniques. A compound which encodes all or part of SEQ ID NO:1 or the reverse complement of a compound which encodes SEQ ID NO:1, and which is at least 18 consecutive base pairs in length is provided as a probe and/or a primer. Preferably, the 18 base pair or more compound is DNA. Most preferred for this use are the DNA compounds which are SEQ ID NO:3, SEQ ID NO:4, and SEQ ID NO:5, or their reverse complements.

Further, this invention provides cells in which the nucleic acid compounds of the invention may be harbored. For example, oocytes wherein nucleic acid compounds of the invention express functional HSmGluR1 receptor are provided. An oocyte wherein DNA expresses functional HSmGluR1 receptor is preferred. Most preferred is an oocyte wherein sense mRNA expresses functional HSmGluR1 receptor.

Other host cells include those which are transfected with a nucleic acid compound which encodes SEQ ID NO:1. The preferred transfected host cells which encode SEQ ID NO:1 are mammalian cells and *E.coli*. Preferred mammalian cells include AV12 cells. Preferred host cells are those which have been transfected

with a recombinant DNA vector. Preferably, the DNA vector comprises SEQ ID NO:2. The most preferred transfected host cells are AV12/pRS121 and *E. coli/*pRS117.

Additionally, the invention provides a method for identifying nucleic acids homologous to a probe of the present invention, which comprises contacting the test nucleic acid with the probe under hybridizing conditions, and identifying nucleic acids which are homologous to the probe. The preferred probes for use in this method are SEQ ID NO:3, SEQ ID NO:4, or SEQ ID NO:5.

Assays utilizing the compounds provided by the present invention are also provided. The assays provided determine whether a substance interacts with or affects the compound SEQ ID NO:1, said assays comprising contacting a functional compound of SEQ ID NO:1 with said substance, monitoring interaction by physically detectable means, and identifying those substances which effect a chosen response.

Preferably, the physically detectable means are competition with labeled glutamate, hydrolysis of phosphatidylinositol (PI), electrophysiological response in an oocyte expression system, stimulation or inhibition of adenylate cyclase or release of arachidonic acid. A most preferred glutamate competition assay utilizes radioisotope-labeled glutamate. A most preferred oocyte expression system utilizes sense mRNA.

The invention also provides a method for constructing a recombinant host cell capable of expressing a nucleic acid compound which encodes a compound which comprises SEQ ID NO:1, said method comprising transfecting a host cell with a recombinant DNA vector that comprises said nucleic acid compound. The preferred method utilizes mammalian cells as the host cells. The most preferred method utilizes AV12 cells as the mammalian host cells. A preferred method includes a DNA vector which comprises SEQ ID NO:2. A most preferred method utilizes the DNA vector pRS121.

Additionally, a method for expressing a nucleic acid sequence which encodes SEQ ID NO:1 in a recombinant host cell is provided. The method comprises culturing a transfected host cell provided by the present invention under conditions suitable for gene expression. The preferred method utilizes mammalian cells as the host cells. The most preferred method utilizes AV12 cells as the mammalian host cells. The more preferred method utilizes a recombinant DNA vector comprising SEQ ID NO:2. The most preferred method utilizes the recombinant DNA vector pRS121.

The following section provides a detailed description of the present invention. For purposes of clarity and as an aid in understanding the invention, as disclosed and claimed herein, the following items are defined below.

"mRNA" - RNA which has been transcribed either in vivo or in vitro, including, for example, RNA transcripts prepared in vitro via transcription of coding sequences of DNA by RNA polymerase.

"Primer" - A nucleic acid fragment or its reverse complement which functions as template for enzymatic or synthetic elongation.

"Probe" - A nucleic acid compound or a fragment thereof, or its reverse complement either of which is used to hybridize to other nucleic acids.

"Part of SEQ ID NO:1" - A sequence containing at least 6 consecutive amino acid residues or more and that corresponds to a sequence contained in SEQ ID NO:1.

"Physically detectable" - Any information which has been presented in humanly recognizable form, with or without the aid of intervening interpretation. For example, electrophysiological, chemical, or biochemical data is considered within the realm of physically detectable information.

"Functional compound of SEQ ID NO:1" - A compound comprising SEQ ID NO:1 which is capable of interacting with glutamate.

"HSmGluR1 receptor" - the amino acid sequence SEQ ID NO:1.

"SEQ ID NO:1 and functional equivalents thereof" - The compound of SEQ ID NO:1 and conserved alterations of the amino acid sequence of SEQ ID NO:1, wherein the conserved alterations result in a compound which exhibits substantially the same physical and structural qualities of SEQ ID:1.

"SEQ ID NO:3" - The DNA sequence ATG GTC GGG CTC CTT TTG TTT TTT CCA GCG ATC TTT TTG GAG GTG TCC CTT CTC CCC.

This sequence includes bases 1 through 60 of SEQ ID NO:2

"SEQ ID NO:4" - CCA GGA CACCTT CTG GAA AAT CCC AAC TTT AAA CGA ATC TGC ACA GGC AAT GAA AGC TTA.

This sequence includes bases 1141 through 1200 of SEQ ID NO.2.

"SEQ ID NO:5" - AAC GTA TCC TAC GCC TCT GTC ATT

This sequence includes bases 3761 through 3817 of SEQ ID NO:2, with the addition of a TAA stop codon at the 3' end.

"Transfection" - any transfer of nucleic acid into a host cell, with or without integration of said nucleic acid into genome of said host cell.

The present invention provides compounds which comprise the amino acid sequence SEQ ID NO:1, and functional equivalents thereof. The preferred amino acid compound is SEQ ID NO:1, which is the following sequence of amino acids:

15	-	Met	Val	Gly	Léu	Leu	Leu	Phe	Phe	Phe	Pro	Ala	Ile	Phe	Leu	Glu	Val
¥.,		Ser	Leu	Leu	Pro	Arg	Ser	Pro	Gly	Arg 25	Lys	Val	Leu	Leu	Ala	Gly	Ala
20	. 4	Ser	Ser	Gln 35	Arg	Ser	Val	Ala	Arg	Met	Asp	Gly	Asp	Val 45	Ile	Ile	Gly

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	Ala	Leu 50	Phe	Ser	Val	His	His 55	Gln _.	Pro	Pro	Ala	Glu 60	Lys	Val	Pro	Glu
5	Arg 65	Lys	Cys	Gly	Gļu	Ile 70	Arg	Glu	Gln	Tyr	Gly 75	Ile	Gln	Arg	Val	Glu 80
X.	Ala	Met	Phe	His	Thr 85	Leu	Asp	Lys	Ile	Asn 90	Ala	Asp	Pro	Val	Leŭ 95	Leu
10	Pro	Asn	Ile	Thr 100		Gly	Ser	Glu	Ile 105		Asp	Ser	Cys	Trp		Ser
15	Ser	Val	Ala 11	Leu	Glu	Gln	Ser	11e 120		Phe	Ile	Arg	Asp 125		Leu	Ile
	Ser	Ile 130		Asp	Glu	Lys	Asp 135		Ile	Asn	Arg	Cys 14		Pro	Asp	Gly
20	Gln 149		Leu	Pro	Pro	Gly 150		Thr	Lys	Lys	Pro 159		Ala	Gly	Val	11e 160
	Gly	Pro	Gly	Ser	Ser 169		Val	Ala	Ile	Gln 170	Val	Gln	Asn	Leu	Leu 179	Gln 5
25	Leu	Phe	Asp	11e 180		Gln	Ile	Ala	Tyr 189		Ala	Thr	Ser	Ile 190	Asp)	Leu
	Ser	Asp	Lys 19		Leu	Tyr	Lys	Tyr 200		Leu	Arg	Val	Val 20	Pro 5	Ser	Asp
3 <i>0</i>	Thr	Leu 210		Ala	Arg	Ala	Met 21	Leu 5	Asp	Ile	Val	Lys 22		Tyr	Asn	Trp
3	Thr 22!	Tyr 5	Val	Ser	Ala	Val 230		Thr	Glu	Gly	Asn 23	Tyr 5	Gly	Glu	Ser	Gly 240
35		Asp			24	5				250	0				25!	5
40		Ser		260	0			•	26	5.	· .		4	27	0	(h
	•	Leu	2.7	5		·		28	0). ·	28	5	•	
45	÷	Phe 29	0				29	5				30	0			
	30	5.				31	0		4		31	5	· .		٠, .	320
50	Ala	Asp	Arg	Asp	G1u 32		Ile	Glu	Gly	Tyr 33	_	Val	Glu	Ala	Asn 33	Gly 5

*	Gly	Ile	Thr	11e 340		Leu	Gln	Ser	Pro 345		Val	Arg	Ser	Phe 350		Asp
5	Tyr	Phe	Leu 355		Leu	Arg	Leu	Asp 360		Asn	Thr	Arg	Asn 365		Trp	Phe
	Pro	Glu 370		Trp	Gln	His	Arg 375	Phe	Gln	Cys	Arg	Leu 380		Gly	His	Leu
10	Leu 385		Asn	Pro	Asn	Phe 390	Lys)	Arg.	Ile	Суз	Thr 395		Asn	Glu	Ser	Leu 400
,-	Glu	Glu	Asn	Tyr	Val 405		Asp	Ser	Lys	Met 410		Phe	Val	Ile	Asn 415	
15	Ile	Tyr	Ala	Met 420		His	Gly	Leu	Gln 425		Met	His	His	Ala 430		Cys
20	Pro	Gly	His 435		Gly	Leu	Cys	Asp. 440		Met	Lys	Pro	Ile 445	_	Gly	Ser
	Lys	Leu 450		Asp	Phe	Leu	Ile 455		Ser	Ser	Phe	Ile 460		Val	Ser	Gly
25	Glu 46		Val	Trp	Phe	Asp 470	Glu)	Lys	Gly.	Asp	Ala 475	_	Gly	Arg	Tyr	Asp 480
*	Ile	Met	Asn	Leu	Gln 485		Thr	Glu	Ala	Asn 490		Tyr	Asp	Tyr	Val-	
30	Val	Gly	Thr	Trp		Glu	Gly	Val	Leu 505		Ile	Asp	Asp	Tyr 510		Ile
*	Gln	Met	Asn 519		Ser	Gly	Val	Val 520		Ser	Val	Cys	Ser 529	Glu 5	Pro	Cys
35	Leu	Lys 53		Gln	Ile	Lys	Val. 535		Arg	Lys	Gly	Glu 540		Ser	Cys	Суз
	Trp		Cys	Thr	Ala	Cys 55	Lys 0	Glu	Asn	Glu	Tyr 555	Val	Gln	Asp.	Glu	Phe 560
40	Thr	Cys	Lys	Ala	Cys 56	_ '	Leu	Gly	Trp	Trp 570		Asn	Ala	Asp	Leu 579	
45	Gly	Cys	Glu	Pro 580		Pro	Val	Arg	Tyr 589	-	Glu	Trp	Ser	Asn 59	Ile O	Glu
	Pro	Ile	Ile 59		Ile	'Ala	Phe	Ser 600		Leu	Gly	Ile	Leu 60		Thr	Leu
						* .		•	٠				*			

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· ;	Phe	Val 610		Leu	Ile	Phe	Val 619		Tyr	Arg	Asp	Thr 620		Val	Val	Lys
5	Ser 625		Ser	Arg	Glu	Leu 630		Tyr	Ile	Ile	Leu 635		Gly	Ile	Phe	Leu 640
	Gly	Tyr	Val	Cys	Pro 645		Thr	Leu	Ile	Ala 65.0	-	Pro	Thr	Thr	Thr 655	
10	Cys	Tyr	Leu	Gln 660	_	Leu	Leu	Val	Gly 665		Ser	Ser	Ala	Met 67(Tyr
1	Ser	Ala	Leu - 675	,	Thr	Lys	Thr	Asn 680	_	Ile	Ala	Arg	Ile 685		Ala	Gly
15	Ser	Lys 690	-	Lys	Ile	Суз	Thr 695	_	Lys	Pro	Arg	Phe 700		Ser	Ala	Trp
20	Ala 705		Va:1	Iļe	Ile	Ala 710		Ile	Leu	Ile	Ser 715		Gln	Leu	Thr	Leu 720
	Val	Val	Thr	Leu	11e 725		Met	Glu	Pro	Pro 730		Pro	Ile	Leu	Ser -735	•
25	Pro	Ser	Ile	Lys 740		Val	Tyr	Leu	Ile 749		Asn	Thr	Ser	Asn 750		Gly
	Val	Val	Ala 755		Leu	Gly	Tyr	Asn 760	-	Leu	Leu	Ile	Met 765	_	Суз	Thr
30	Tyr	Tyr 770		Phe	Lys	Thr	Arg 779		Val	Pro	Ala	Asn 780		Asn	Glu	Ala
1	Lys 785		Ile	Ala	Phe	Thr 790		Tyr	Thr	Thr	Cys 795		Ile	Trp	Leu	Ala 800
35	Phe	Val	Pro	Ile.	Tyr 805		Gly	Ser	Asn	Tyr 810	_	Ile	Ile	Thr	Thr 815	
131	Phe	Ala	Val	Ser 820		Ser	Val	Thr	Val 825		Leu	Gly	Cys	Met - 830		Thr
40	Pro	Lys	Met 835	-	Ile	Ile	Ile	Ala 840		Pro	Glu	Arg	Asn 849	_	Arg	Ser
45	Ala	Phe 850		Thr	Ser	Asp	Val 859	_ `	Arg	Met	His	Val . 860		Asp	Gly	Lys
	Leu 869	Pro	Cys	Arg	Ser	Asn 870	_	Phe	Leu	Asn	Ile 875		Arg	Arg	Lys	Lys 880
50	Ala	Gly	Ala	Gly	Asn 885		Asn	Ser	Asn	Gly 890		Ser	Val	Ser	Trp 895	
*		•													•	

•																
	Glu I	Pro	Gly	Gly 900		Gln	Val		Lys 905		Gln	His	Met	Trp 910	His	Arg
5	Leu S	Ser	Val 915		Val	Lys	Thr	Asn 920		Thr	Ala	Cys	Asn 929		Thr	Ala
*	Val :	11e 930		Pro	Leu	Thr	Lys 935		Tyr	Gln	Gly	Ser 940		Lys	Ser	Leu
. 10	Thr 1	Phe	Ser	Asp.	Thr	Ser 950		Lys	Thr	Leu	Tyr 955		Val	Glu	Glu	Glu 960
· · · · · · · · · · · · · · · · · · ·	Glu i	Asp	Ala	Gln	Pro 965		Arg	Phe	Ser	Pro 970		Gly	Ser	Pro	Ser 975	Met
15	Val V	Val	His	Arg 980		Val	Pro	Ser	Ala 985		Thr	Thr	Pro	Pro 990	Leu)	Pro
	Pro l	His	Leu 995		Ala	Glu	Glu	Thr		Leu	Phe	Leu	Ala 100		Pro	Ala
20	Leu	Pro 101		Gly	Leu	Pro	Pro.		Leu	Gln	Gln	Gln 102		Gln	Pro	Pro
25	Pro (Gln	Lys	Ser	Leu 10:	Met 30	Asp	Gln	Leu	Gln 103	Gly 5	Val	Val	Ser	Asn 1040
s **	Phe	Ser	Thr	Ala	Ile 104		Asp	Phe	Hiş	Ala 109	Val	Leu	Ala	Gly	Pro 105	Gly 55
30	Gly	Pro	Gly	Asn 106		Leu	Arg	Ser	Leu 10		Pro	Pro	Pro	Pro 107	Pro	Pro
*	Gln	His	Leu 107		Met	Leu	Pro	Leu 108		Leu	Ser	Thr	Phe 10	Gly 85	Glu	Glu
35	Leu	Val 109		Pro	Pro	Ala	Asp		Asp	Asp	Asp	Ser		Arg	Phe	Lys
* * *	Leu 110		Gln	Glu	Tyr	Val		Glu	His	Glu	Arg		Gly	Asn	Thr	Glu 1120
40	Glu	Asp	Glu	Leu	Glu 11		Glu	Glu	Glu	Asp		Gln	Ala	Ala	Ser	Lys 35
	Leu	Thr	Pro	Asp		Ser	Pro	Ala	Leu 11		Pro	Pro	Ser	Pro		Arg
45	Asp	Ser	Val		Ser	Gly	Ser	Ser		Pro	Ser	Ser	Pro	Val 65	Ser	Glu
50					*			•				* / *				- •
***	Ser	Val 11		Cys	Thr	Pro	Pro		Val	Ser	Tyr	Ala 11	Ser 80	Val	Ile	Leu
55	Arg		Tyr	Lys	Gln		- Ser 90	Ser	Thr	Leu		,**				

Those in the art will recognize that some alterations of SEQ ID NO:1 will fail to change the function of the amino acid compound. For instance, some hydrophobic amino acids may be exchanged for other hydrophobic amino acids. Those altered amino acid compounds which confer substantially the same function in substantially the same manner as the exemplified amino acid compound are also included in the present invention.

Artisans will also recognize that SEQ ID NO:1 and functional equivalents thereof may be synthesized by a number of different methods. All of the amino acid compounds of the invention can be made by chemical methods well known in the art, including solid phase peptide synthesis or recombinant methods. Both methods are described in U.S. Patent 4,617,149. Recombinant methods are preferred if a high yield is desired. A general method for the construction of any desired DNA sequence is provided in Brown *et al.*, 68 Methods in *Enzymology* 109 (1979).

Other routes of producing the amino acid compounds are well known. Expression in eucaryotic cells can be achieved via SEQ ID NO:2. For example, the amino acid compounds can be produced in eucaryotic cells using SV40-derived expression vectors comprising DNA which encodes for SEQ ID NO:1. Some viruses are also appropriate vectors for this purpose. For example, the adenovirus, the adeno associated virus, the vaccinia virus, the herpes virus, the baculovirus, and the Rous sarcoma virus are useful viral vectors. Such a method is described in U.S. Patent 4,775,624. Several alternate methods of expression are described in J. Sambrook, E.F. Fritsch & T. Maniatis, *Molecular Cloning: A Laboratory Manual* 16.3-17.44 (1989).

Other embodiments of the present invention are nucleic acid compounds which comprise nucleic acid sequences which encode SEQ ID NO:1. As those in the art will recognize, the amino acid compounds of the invention can be encoded by a multitude of different nucleic acid sequences because most of the amino acids are encoded by more than one nucleic acid triplet. Because these alternate nucleic acid sequences would encode the same amino acid sequences, the present invention further comprises these alternate nucleic acid sequences. Preferably, the nucleic acid compound is DNA or sense mRNA. A most preferred embodiment of a DNA compound encoding an HSmGluR1 receptor has this sequence:

£	ATGGTCGGGC	TCCTTTTGTT	TTTTTTCCCA	GCGATCTTTT	TGGAGGTGTC	CCTTCTCCCC	60
0	AGAAGCCCCG	GCAGGAAAGT	GTTGCTGGCA	GGAGCGTCGT	CTCAGCGCTC	GGTGGCCAGA	120
	ATGGACGGAG	ATGTCATCAT	TGGAGCCCTC	TTCTCAGTCC	ATCACCAGCC	TCCGGCCGAG	180
· ·	AAAGTGCCCG	AGAGGAAGTG	TGGGGAGATC	AGGGAGCAGT	ATGGCATCCA	GAGGGTGGAG	240
э·	GCCATGTTCC	ACACGTTGGA	TAAGATCAAC	GCGGACCCGG	TCCTCCTGCC	CAACATCACC	300
	CTGGGCAGTG	AGATCCGGGA	CTCCTGCTGG	CACTCTTCCG	TGGCTCTGGA	ACAGAGCATT	360
O .	GAGTTCATTA	GGGACTCTCT	GATTTCCATT	CGAGATGAGA	AGGATGGGAT	CAACCGGTGT	420
	CTGCCTGACG	GCCAGTCCCT	CCCCCAGGC	AGGACTAAGA	AGCCCATTGC	GGGAGTGATC	480
	GGTCCCGGCT	CCAGCTCTGT	AGCCATTCAA	GTGCAGAACC	TGCTCCAGCT	CTTCGACATC	540
5	CCCCAGATCG	CTTATTCAGC	CACAAGCATC	GACCTGAGTG	ACAAAACTTT	GTACAAATAC	600

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	TTCCTGAGGG	TTGTCCCTTC	TGACACTITG	CAGGCAAGGG	CCATGCTTGA	CATAGTCAAA	660
5	CGTTACAATT	GGACCTATGT	CTCTGCAGTC	CACACGGAÁG	GGAATTATGG	GGAGAGCGGA	720
	ATGGACGCTT	TCAAAGAGCT	GGCTGCCCAG	GAAGGCCTCT	GTATCGCCCA	TTCTGACAAA	78
	ATCTACAGCA	ACGCTGGGGA	GAAGAGCTTT	GACCGACTCT	TGCGCAAACT	CCGAGAGAGG	840
10	CTTCCCAAGG	CTAGAGTGGT	GGTCTGCTTC	TGTGAAGGCA	TGACAGTGCG	AGGACTCCTG	900
• •	AGCGCCATGC	CCCCCTTCC	CGTCGTGGGC	GAGTTCTCAC	TCATTGGAAG	TGATGGATGG	9.60
*	GCAGACAGAG	ATGAAGTCAT	TGAAGGTTAT	GAGGTGGAAG	CCAACGGGGG	AATCACGATA	1020
15	AAGCTGCAGT	CTCCAGAGGT	CAGGTCATTT	GATGATTATT	TCCTGAAACT	GAGGCTGGAC	1080
•	ACTAACACGA	GGAATCCCTG	GTTCCCTGAG	TTCTGGCAAC	ATCGGTTCCA	GTGCCGCCTT	1140
	CCAGGACACC	TTCTGGAAAA	TCCCAACTTT	AAACGAATCT	GCACAGGCAA	TGAAAGCTTA	1200
20	GAAGAAAACT	ATGTCCAGGA	CAGTAAGATG	GGGTTTGTCA	TCAATGCCAT	CTATGCCATG	1260
	GCACATGGGC	TGCAGAACAT	GCACCATGCC	стстсссстс	GCCACGTGGG	CCTCTGCGAT	1320
	GCCATGAAGC	CCATCGACGG	CAGCAAGCTG	CTGGACTTCC	TCATCAAGTC	CTCATTCATT	1380
25	GGAGTATCTG	GAGAGGAGGT	GTGGTTTGAT	GAGAAAGGAG	ACGCTCCTGG	AAGGTATGAT	1440
	ATCATGAATC	TGCAGTACAC	TGAAGCTAAT	CGCTATGACT	ATGTGCACGT	TGGAACCTGG	1500
	CATGAAGGAG	TGCTGAACAT	TGATGATTAC	AAAATCCAGA	TGAACAAGAG	TGGAGTGGTG	1560
30 	CGGTCTGTGT	GCAGTGAGCC	TTGCTTAAAG	GGCCAGATTA	AGGTTATACG	GAAAGGAGAA	1620
	GTGAGCTGCT	GCTGGATTTG	CACGGCCTGC	AAAGAGAATG	AATATGTGCA	AGATGAGTTC	1680
35	ACCTGCAAAG	CTTGTGACTT	GGGATGGTGG	CCCAATGCAG	ATCTAACAGG	CTGTGAGCCC	/1740
	ATTCCTGTGC	GCTATCTTGA	GTGGAGCAAC	ATCGAACCCA	TTATAGCCAT	CGCCTTTTCA	1800
	TGCCTGGGAA	TCCTTGTTAC	CTTGTTTGTC	ACCCTAATCT	TTGTACTGTA	CCGGGACACA	1860
40	CCAGTGGTCA	AATCCTCCAG	TCGGGAGCTC	TGCTACATCA	TCCTAGCTGG	CATCTTCCTT	1920
	GGTTATGTGT	GCCCATTCAC	TCTCATTGCC	AAACCTACTA	CCACCTCCTG	CTACCTCCAG	1980
	CGCCTCTTGG	TTGGCCTCTC	CTCTGCGATG	TGCTACTCTG	CTTTAGTGAC	TAAAACCAAT	2040
45	CGTATTGCAC	GCATCCTGGC	TGGCAGCAAG	AAGAAGATCT	GCACCCGGAA	GCCCAGGTTC	2100
• •	ATGAGTGCCT	GGGCTCAGGT	GATCATTGCC	TCAATTCTGA	TTAGTGTGCA	ACTAACCCTG	2160

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	GTGGTAACCC	TGATCATCAT	GGAACCCCCT	ATGCCCATTC	TGTCCTACCC	AAGTATCAAG	2220
*	GAAGTCTACC	TTATCTGCAA	TACCAGCAAC	CTGGGTGTGG	TGGCCCCTTT	GGGCTACAAT	2280
5	GGACTCCTCA	TCATGAGCTG	TACCTACTAT	GCCTTCAAGA	CCCGCAACGT	GCCCGCCAAC	2340
•	TTCAACGAGG	CCAAATATAT	CGCGTTCACC	ATGTACACCA	CCTGTATCAT	CTGGCTAGCT	2400
	TTTGTGCCCA	TTTACTTTGG	GAGCAACTAC	AAGATCATCA	CAACTTGCTT	TGCAGTGAGT	2460
10	CTCAGTGTAA	CAGTGGCTCT	GGGGTGCATG	TTCACTCCCA	AGATGTACAT	CATTATTGCC .	2520
•	AAGCCTGAGA	GGAATGTCCG	CAGTGCCTTC	ACCACCTCTG	ATGTTGTCCG	CATGCATGTT	2580
15	GGCGATGGCA	AGCTGCCCTG	CCGCTCCAAC	ACTTTCCTCA	ACATCTTCCG	AAGAAAGAAG	2640
	GCAGGGGCAG	GGAATGCCAA	TTCTAATGGC	AAGTCTGTGT	CATGGTCTGA	ACCAGGTGGA	2700
	GGACAGGTGC	CCAAGGGACA	GCATATGTGG	CACCGCCTCT	CTGTGCACGT	GAAGACCAAT	2760
20	GAGACGGCCT	GCAACCAAAC	AGCCGTCATC	AAACCCCTCA	CTAAAAGTTA	CCAAGGCTCT	2820
	GGCAAGAGCC	TGACCTTTTC	AGATACCAGC	ACCAAGACCC	TTTACAACGT	AGAGGAGGAG	2880
•	GAGGATGCCC	AGCCGATTCG	CTTTAGCCCG	CCTGGTAGCC	CTTCCATGGT	GGTGCACAGG	2940
25	CGCGTGCCAA	GCGCGGCGAC	CACTCCGCCT	CTGCCGCCCC	ACCTGACCGC	AGAGGAGACC	3000
	CCCCTCTTCC	TGGCCGAACC	AGCCCTCCCC	AAGGCTTGC	CCCCTCCTCT	CCAGCAGCAG	3060
	CAGCAACCCC	CTCCACAGCA	GAAATCGCTG	ATGGACCAGC	TCCAGGGAGT	GGTCAGCAAC	3120
30	TTCAGTACCG	CGATCCCGGA	TTTTCACGCG	GTGCTGGCAG	GCCCCGGGG	TCCCGGGAAC	3180
	GGCTGCGGT	CCCTGTACCC	GCCCCGCCA	CCTCCGCAGC	ACCTGCAGAT	GCTGCCGCTG	3240
35	CAGCTGAGCA	CCTTTGGGGA	GGAGCTGGTC	TCCCCGCCCG	CGGACGACGA	CGACGACAGC	3300
	GAGAGGTTTA	AGCTCCTCCA	GGAGTACGTG	TATGAGCACG	AGCGGGAAGG	GAACACCGAA	3360
	GAAGACGAAC	TGGAAGAGGA	GGAGGAGGAC	CTGCAGGCGG	CCAGCAAACT	GACCCCGGAT	3420
40	GATTCGCCTG	CGCTGACGCC	TCCGTCGCCT	TTCCGCGACT	CCCTCCCCTC	GGGCAGCTCG	3480
	GTGCCCAGCT	CCCCAGTGTC	CGAGTCGGTG	CTCTGCACCC	CTCCCAACGT	ATCCTACGCC	3540
	TCTGTCATTC	TGCGGGACTA	CAAGCAAAGC	TCTTCCACCC	TG		: "

This is the sequence identified as SEQ ID NO:2.

E. coli/pRS117, which contains a cloning vector comprising SEQ ID NO:2, was deposited and made part of the stock culture collection of the Northern Regional Research Laboratories (NRRL), Agricultural Research Service, U.S. Department of Agriculture, Peoria, Illinois, 61604 on April 22, 1992, under the accession number NRRL B-18969. SEQ ID NO:2 can be isolated from the plasmid, for example, as a 4.1 kb EcoRI restriction fragment. Other fragments may also be useful in obtaining SEQ ID NO:2.

Additionally, the DNA sequences can be synthesized using automated DNA synthesizers, such as the ABS (Applied Biosystems, 850 Lincoln Centre Drive, Foster City, CA 94404) 380B DNA synthesizer. The DNA sequences can also be generated by the polymerase chain reaction (PCR) as described in U.S. Patent No. 4,889,818.

As art workers will recognize, many vectors are available for expression and cloning. Those expression and cloning vectors which comprise nucleic acids which encode SEQ ID NO:1 are included in the present invention. Preferred nucleic acid vectors are those which are DNA. A most preferred recombinant DNA

vector comprises the DNA sequence SEQ ID NO:2. Plasmid pRS117 is a preferred DNA vector of the present invention.

Other preferred DNA vectors include those which comprise a promoter positioned to drive expression of SEQ ID NO:2. Preferred expression vectors include those which function in mammalian cells. Preferred mammalian expression vectors include those which function in AV12 cells. Most preferred for expression in AV12 cells is the expression vector pRS121.

Restriction fragments of these vectors are also provided. The preferred fragments are the 4.1 kb EcoRl restriction fragment and the 3.8 kb BssHll/AfIII restriction fragment of plasmid pRS117.

Plasmid pRS117 may be isolated from the deposited *E. coli*/pRS117, using an ordinary cesium chloride DNA isolation procedure. Plasmid pRS117 is readily modified to construct expression vectors that produce HSmGluR1 receptors in a variety of organisms, including, for example, *E. coli*, Sf9 (as host for baculovirus), *Spodoptera* and *Saccharomycetes*. The current literature contains techniques for constructing AV12 expression vectors and for transfecting AV12 host cells. For example, U.S. Patent No. 4,992,373 explains these techniques.

The construction protocols utilized for AV12 vectors can be followed to construct analogous vectors for other organisms, merely by substituting, if necessary, the appropriate regulatory elements using well known techniques. Promoters which may be used, for example, are the thymidine kinase promoter, the metal-lothionin promoter or various viral and immunoglobulin promoters.

The present invention also provides probes and primers useful for molecular biology techniques. A compound which encodes all or part of SEQ ID NO:1, or the reverse complement of a compound which encodes SEQ ID NO:1, and which is at least 18 consecutive base pairs in length is provided as a probe and/or a primer. Preferred probes and primers are DNA. Most preferred probes and primers are: SEQ ID NO:3, SEQ ID NO:4, and SEQ ID NO:5. The techniques associated with using probes and primers are well known in the art.

Any sequence of at least 18 base pairs in length of the nucleic acids of the present invention may be used to screen any other nucleic acid. For example, 18 consecutive bases or more of nucleic acids of the present invention may be used to hybridize to the terminal ends of the coding sequence. Then, through polymerase chain reaction amplification, the full length sequence may be generated. The full length sequence can be subsequently subcloned into any vector of choice.

Alternatively, SEQ ID NO:3, SEQ ID NO:4, or SEQ ID NO:5 may be radioactively labeled in order to screen cDNA libraries by conventional means. Furthermore, any piece of HSmGluR1 DNA which has been bound to a filter may be flooded with total mRNA transcripts, in order to then reverse-transcribe the mRNA transcripts which bind.

Primers and probes may be obtained by means well known in the art. For example, once pRS117 is isolated, restriction enzymes and subsequent gel separation may be used to isolate the fragment of choice.

Host cells which harbor the nucleic acids of the present invention are also provided. For example, occytes which have been injected with RNA or DNA compounds of the present invention are provided. Most preferred occytes of the present invention are those which harbor sense mRNA. Other preferred host cells include AV12 and *E. coli* cells which have been transfected with a vector which comprises SEQ ID NO:2. Most preferred AV12 and *E. coli* host cells are AV12/pRS121 and *E. coli*/pRS117.

The oocyte expression system can be constructed according to the procedure described in Lübbert, et al. 84 Proc. Nat. Acad. Sci. 4332 (1987) or Berger, Methods in Enzymology, Vol. 152 (1987). Other host cell transfection methods are well known in the art as well.

The present invention also provides a method for constructing a recombinant host cell capable of expressing SEQ ID NO:1, said method comprising transfecting a host cell with a recombinant DNA vector that comprises an DNA sequence which encodes SEQ ID NO:1.

The preferred host cell is AV12, which may be obtained from the American Type Culture Collection (ATCC) 12301 Parklawn Drive, Rockville, MD 20852 under the accession number ATCC CRL 9595. The preferred vector for expression is one which comprises SEQ ID NO:2. Especially preferred for this purpose is pRS121.

Other preferred host cells for this method are mammalian cells. Especially preferred mammalian cells are AV12 cells. A preferred AV12 expression vector is pRS121. Transfected host cells may be cultured under well known conditions such that SEQ ID NO:1 is expressed, thus producing HSmGluR1 activity in the recombinant host cell.

Therefore, also provided by the present invention is a method for expressing a gene which encodes SEQ ID NO:1 in a recombinant host cell, said method comprising culturing said transfected host cell under conditions suitable for gene expression. A preferred method utilizes mammalian cells. A preferred method utilizes AV12 cells as the mammalian cells. A most preferred method utilizes AV12 cells as host cells for

pRS121. Expression in host cells may be accomplished according to the procedures outlined in Goeddel, *Methods in Enzymology*, vol. 185 (1990).

Additionally, the invention provides a method for identifying nucleic acids homologous to a probe of the present invention, which comprises contacting a test nucleic acid with the probe under hybridizing conditions and identifying those test nucleic acids which are homologous to the probe. The preferred probes for use in this method are SEQ ID NO:3, SEQ ID NO:4, or SEQ ID NO:5. Hybridization techniques are well known in the art. Sambrook, et al., Molecular Cloning: A Laboratory Manual 11 (1989) describe such procedures.

Assays utilizing the compounds provided by the present invention are also provided. These assays determine whether a substance interacts with or affects the compound of SEQ ID NO:1, said assay comprising contacting a functional compound of SEQ ID NO:1 with said substance, monitoring interaction by physically detectable means, and identifying those substances which effect a chosen response.

Preferably, the physically detectable means are competition with labeled glutamate, hydrolysis of phosphatidylinositol, electrophysiological response in an oocyte expression system, stimulation or inhibition of adenylate cyclase or release of arachidonic acid. A most preferred glutamate competition assay utilizes radioisotope-labeled glutamate. A most preferred oocyte expression system utilizes sense mRNA.

The occyte expression system can be constructed according to the procedure described in Lübbert, et al. 84 Proc. Nat. Acad. Sci. 4332 (1987) or Berger, Methods in Enzymology, Vol. 152 (1987). The radiolabeled HSmGluR1 competition assay may be accomplished according to Foster and Fagg, 7 Brain Res. Rev. 103 (1984). The assay which measures glutaminergic activity via phosphatidylinositol hydrolysis may be accomplished according to Berridge M., 212 Biochem. J. 849 (1983) or Schoepp et al., 11 TiPS 508 (1990). Stimulation and inhibition of adenylate cyclase may be accomplished according to Nakajima et al., 267 J. Biol. Chem. 2437 (1992). Measurement of arachidonic acid release may be accomplished according to Felder et al., 264 J. Biol. Chem. 20356 (1989). Skilled artisans will recognize that desirable K₁ values are dependent on the selectivity of the compound tested. For example, a compound with a K₁ which is less than 10 nM is generally considered an excellent candidate for drug therapy. However, a compound which has a lower affinity, but is selective for the particular receptor, may be an even better candidate. The present invention, however, provides competition assays, which indicate whether a substance has either a high affinity or low affinity to HSmGluR1 receptor, because skilled artisans will recognize that any information regarding binding or selectivity of a particular compound is beneficial in the pharmaceutical development of drugs.

The following are examples of the present invention:

Example 1

Growth of E.coli/pRS117

A lyophilized culture of *E. coli* containing plasmid pRS117 can be obtained from the NRRL, Peoria, Illinois, 61604, under the accession number NRRL B-18969, and inoculated into a suitable broth for the growth of *E. coli* using standard microbiological procedures.

The contents of a lyophil vial containing *E. coli*/pRS117 were transferred into 100 ml of sterile YT (tryptone-yeast extract) broth containing 100 µg/ml ampicillin in a one liter fermentation flask and shaken at 37 °C on an orbital shaker at 250-300 rpm. After the optical density (OD, measured at 600 millimicrons) had reached approximately 1-2 OD, the bacterial cells were recovered and used for the isolation of plasmid pRS117 according to the procedures detailed in J. Sambrook *et al.*, *Molecular Cloning*, Chapter 1, (1989).

Once isolated from the bacterial cells, the plasmid DNA served as a source for the DNA encoding the HSmGluR1 receptor protein. One convenient method to remove the receptor-encoding DNA from plasmid pRS117 was to digest the plasmid with the restriction enzyme EcoRI. This enzyme cuts the plasmid at unique sites to produce a DNA fragment of approximately 4.1 kb containing the entire coding sequence of the HSmGluR1 receptor.

Example 2

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Construction of pRS121 from pRS117

DNA encoding SEQ ID NO:1 was recovered from plasmid pRS117 as described in Example 1. DNA linkers were added to the fragment ends in order to adapt the EcoRI cohesive termini into Bam HI cohesive termini. This was accomplished by ligation of short dupl x oligonucleotides having both an EcoRI terminus

and a BamHI terminus to the pRS117-derived fragments. The ligation involved incubation with T4 DNA ligase.

Following incubation, the reaction products were digested with BamHI and separated according to molecular weight on an agarose gel. The DNA band on the gel at the position expected for a fragment of approximately 4 kb was excised from the gel and the DNA recovered by the phenol-freeze-fracture method of Huff et al., 10 Biotechniques 724 (1991).

The isolated fragment was then ligated to a modified pHD plasmid. The modified pHD vector was substantially the vector described in issued Patent No. 4,992,373, except that the vector described in the issued patent was digested with the restriction enzyme BcII and treated with alkaline phosphatase. The ligation products were then transfected into *E.coli* DH5 α cells which had been made competent for DNA transfection. These cells were plated at low density on TY agar plates which contained ampicillin.

A clone was selected from the colonies which grew. This clone, pRS121 was characterized by restriction enzyme digestion and DNA hybridization probing.

Example 3

Transfection and Growth of AV12/pRS121

AV12 cells were grown in a routine manner. Cells were placed in 100 mm cell-culture plates in Dulbecco's modified Eagle's medium (D-MEM) which contained 10% fetal calf serum at 37 °C and grown in an atmosphere containing 5% CO₂. To prepare plasmid DNA for transfection, plasmid pRS121 (20 µg) was added to 500 µl of 0.5M CaCl₂ and mixed with 500µl 0.9% NaCl buffered at pH 6.95 with N,N-bis(2-hydroxyethyl)-2-aminoethanesulfonic acid. After 30 minutes at room temperature, the this mixture resulted in a suspension of precipitated DNA.

The suspension of precipitated DNA was then added to a 100 mm diameter cell-culture plate of AV12 cells. At the time of adding the suspension of DNA, the cell monolayer was approximately 50% confluent. The plate was incubated overnight at 37°C and, after rinsing with D-MEM, the cells were incubated an additional 24 hours in D-MEM containing 10% dialyzed fetal calf serum. The cells were then detached from the plate with trypsin and dispensed into 10 new plates. After an additional 24 hours incubation, hygromycin, at 200 µg/ml final concentration, was added to the growth medium in order to select for colonies of cells which grew. Those cells which grew contained pRS121 with the associated hygromycin resistance gene. After the clones had reached a suitable size, (1-2 mm in diameter), individual clones were recovered and cultures of these clones were propagated using standard tissue-culture techniques. The cultures were grown in a routine manner in D-MEM medium without glutamic acid, containing 2 mM glutamine and 10% dialyzed fetal calf serum. Subcultures were prepared when cultures became confluent. The subculture preparation included disassociating the cells with trypsin, diluting the cells disassociated into fresh culture medium, and placing the dilutions into fresh culture vessels at 1/10 the original concentration.

Example 4

AV12/pRS121 Pl Assay

PI hydrolysis in clonal cell lines of AV12/pRS121 cells was measured in response to glutamate agonists as a functional assay for metabotropic glutamate receptor activity according to Schoepp, 11 *TiPS* 508 (1990).

Twenty-four-well tissue-culture vessels were seeded with approximately 250,000 cells per well in D-MEM (in the absence of glutamic acid) which contained 2 mM glutamine and 10% dialyzed fetal calf serum. Four microcuries of ³H-myoinositol were then added to each well and the cultures were incubated for 48 hours at 37 °C. The wells were then rinsed with serum-free medium which contained 10mM LiCl and 10mM myoinositol. Some wells were then exposed to medium with glutamate agonists for one hour at 37 °C, and some wells were at the same time exposed to medium without glutamate agonists for one hour at 37 °C.

The reactions were terminated by removing the media and adding 0.5 ml acetone-methanol (1:1). The cells were then incubated at 4 °C for 20 minutes. The acetone-methanol solutions were recovered from the wells and were placed into centrifuge tubes. Each well was rinsed with 0.5 ml water and the rinses were combined in the centrifuge tubes with the corresponding solvent extracts. After centrifugation at 15,000g for 10 minutes, the supernatants were recovered.

In order to separate the PI hydrolysis products, ACELL PLUS QMA (Waters Division of Millipore Corporation) cartridges were prepared by adding 10 ml of a solution containing a final concentration of 1M

'ammonium formate and 0.1 M formic acid to each cartridge, followed by two rinses with 10 ml distilled water. The cell extracts were were diluted to 5 ml with distilled water and were then added to individual cartridges. Each cartridge was then washed with 10 ml of 5 mM sodium tetraborate solution.

Labeled phosphoinositides were eluted from the cartridges with 4 ml of a solution which contained: 0.1 M ammonium formate; 0.1 M formic acid and 5 mM sodium tetraborate. The eluates were collected in scintillation vials. Scintillation counting fluid (Ready Solv HP) was added to the vials and the radioactivity was determined in a Beckmann Scintillation Counter.

Exposure of the AV12/pRS121 to the known glutamate receptor agonist quisqualate (10µM) resulted in a 200 to 300 percent increase in PI hydrolysis over the basal level in cells with exposure to agonist. No increase in PI hydrolysis over basal levels was found in control AV12 cells.

Example 5

In vitro transcription of RNA sing pRS117 as a DNA template

RNA transcripts encoding the HSmGluR1 receptor were produced by enzymatic transcription from pRS117 using an RNA polymerase which recognizes the transcription promoter contained in the plasmid adjacent to the amino terminal coding end of the receptor subunit cDNA. Plasmid pRS117 was treated with the restriction enzyme Sall which made a single cut distal to the 3' end of the cDNA insert in the circular DNA and converted the plasmid DNA into a linear form. This DNA was then incubated with T7 RNA polymerase in the presence of GpppG cap nucleotide, rATP, rCTP, rUTP and rGTP. The synthetic RNA transcript obtained was purified by passage over a Sephadex G-50 column. For a detailed description of *in vitro* RNA synthesis using bacteriophage RNA polymerase such as T7, see P. A. Krieg and D. A. Melton, Vol 155, *Methods in Enzymology*, Ch. 25, 1987.

Example 6

Functional Expression of HSmGluR1 Receptor in Xenopus Oocytes.

Oocytes suitable for injection were obtained from the adult female *Xenopus laevis* using procedures described in C. J. Marcus-Sekura and M. J. M. Hitchcock, *Methods in Enzymology*, Vol. 152 (1987). After treatment with collagenase type 1a (Sigma) at a concentration of 2 mg/ml, the defolliculated oocytes were injected essentially as described by M. J. M. Hitchcock *et al.*, *Methods in Enzymology*, Vol. 152 Chapter 28, (1987). Subsequently, 5 ng of RNA transcript in a total volume of 50 nl, prepared as described in Example 2, were injected into each oocyte and they were then incubated in Barth's saline solution at 18 °C until needed for electrophysiological measurements.

In order to detect the presence of HSmGluR1 receptor, the ability of the receptor to function was determined by voltage recording of electrical current flowing across the oocyte membrane in response to exposure to glutamate agonists. Individual occytes were placed in a diffusion chamber (0.5 ml vol.) through which solutions were perfused rapidly. Drugs (agonists and antagonists) were applied to the oocytes by adding them to the perfusing solutions and subsequently washing them out with control solution. The control solution contained 96 nM NaCl, 2mM KCl, 1.8 nM CaCl2, 1 mM MgCl2, and 5 mM HEPES buffer, pH 7.6. After insertion of electrodes into the oocytes, voltage recordings were made using the bridge circuit of an Axoclamp 1A voltage-clamp unit. Microelectrodes were filled with 3 M CsCl. Electrophysiological recordings of the oocytes clamped at -70 mV were made at room temperature (20-25 °C), 3 days or more after injection of RNA into the oocytes. In response to perfusion of the oocytes with 10 µM glutamate, an inward current across the oocyte membrane of 400 nano-amperes was observed. The current observed was proportional to the concentration of agonist in the perfusion fluid. From the values obtained, EC50 values (the concentration at which 50% of maximal response was observed) were calculated for various agonists. For example, the EC50 value for glutamate was 0.000002 M and the EC50 value for quisqualate was 0.0000004 M. As those skilled in the art appreciate these results are indicative of a metabotropic glutamate receptor. For a detailed discussion of the electrophysiology of Xenopus oocytes see N. Dascal, 22 CRC Critical Reviews in Biochemistry, 317 (1987).

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	:	•	Gln	Met	Asn 515	Lys	Ser	Gly	Val	'Val 520	Arg	.Ser	Val	Суз	Ser 525	Glu	Pro	Су
5			Leu	Lys 530	Gly	Gln	Ile	Lys	Val 535	Ile	Arg	Lys	Gly	G1ú 540	Val	Ser	Cys	Сув
			Trp 545	Ile	Cys	Thr	λla	Cys 550	_	Glu	λsn	Glu	Tyr 555	Val	Gln	λsp	Glu	Phe 560
0		*	Thr	Сув	Lys	Ala	Сув 565	yab	Leu	Gly	Trp	Trp -570	Pro	λsn	Ala	Авр	Leu 575	Thr
		: :	Gly	Сув	Ģlu	Pro 580	Ile	Pro	Val	Arg	Tyr 585	Leu	Glu	Trp	Ser	As n 590	Ile	Glu
5	· · · · · · · · · · · · · · · · · · ·		Pro		Ile 595	Ala	Ile	Ala	Phe	Ser 600	Сув	Leu	Gly	Il•	Leu 605	Val	Thr	Leu
			Phe	Val 610	Thr	Leu	Ile	Phe	Val. 615	Leu	Tyr	Arg	λsp	Thr 620	Pro	Val	Val	Lys
0			Ser 625	Ser	Ser	Arg	Glu	Leu 630	Сув	Tyr	Ile	Ile	Leu 635	Ala	Gly	Ile	Phe	Leu 640
	-	·	Gly	Tyr	Val	Cys	Pro 645		Thr	Leu	Įle	Ala 650	Lys	Pro	Thr	Thr	Thr 655	Ser
	•	•	Cys	Tyr	Leu	Gln 660	Arg	Leu	Leu	Val	Gly 665	Leu	Ser	Ser	Ala	Met 670	Сув	Tyr
?5			Ser	Ala	Leu 675	Va1	Thr	Lys	Thr	Asn 680	Arg	I1•	Ala	Arg	11e 685	Leu	Ala	Gly
		•	Ser	Lys 690	Lys	Lys	Ile	Сув	Thr 695	Arg	Lys	Pro	Arg	Phe 700	Met	Ser	Ala	.Trp
30	·		Ala 705	Gln	Val	Ile	Ile	Ala 710	Ser	Ile	Leu	Ile	Ser 715		Gln	Leu	Thr	Leu 720
	· · · · ·		Va1	Val	Thr		Ile 725		Met	Glu	Pro	Pro 730	Met	Pro	Ile	Leu	Ser 735	Tyr
35	1		Pro	Ser	Ile	Lys 740	Glu	Val	Tyr	Leu	Ile 745	Cys	Asn	Thr	Ser	Asn 750	Leu	Gly
· ·	• • • • •		Val	Val	Ala 755	Pro	Leu	G1y	Tyr	A sn 760	Cly	Leu	Leu	Ile	Met 765	Ser	Сув	Thr
10	∞		-	Tyr 770	Ala	Phe	Lys	Thr	Arg 775	Asn	Val	Pro	Ala	As n 780	Phe.	λsn	Glu	Ala
	ē.		Lys 785	Tyr	Ile	Ala	Phe	Thr 790	Met	Туг	Thr	Thr	Cys 795	I1•	Ile	Trp	Leu	Ala 800
			Phé	Val	Pro	Ile	Tyr 805	Phe	Gly	Ser	λsn	Tyr 810	Lys	Il•	Ile	Thr	Thr 815	Сув
15	11.		Phe	Ala	Val	Ser 820	Leu	Ser	Val	Thr	Val 825	Ala	Leu	Gly	Cýs	Met 830	Phe	Thr
*			Pro	Lys	Met 835	Tyr	Ile	Ile	Ile	Ala 840	Lys	Pro	Glu	Arg	Asn 845	Val	Arg	Ser
50		··.	Ala	Phe	Thr	Thr	Ser	λвр	Val	Val	Arg	Het	His	Val	Gly	yab	Gly	Lys

•		850					855					860	, j	*		.:
	Leu	Pro	Cys	Aro	Ser	. Asn	Thr	Phe	Leu	ı Asn	Ile	Phe	Arc	. Aro	i Ive	; L T.VE
· -	865		-2-			870					875	_		, ,,,,,	, Ly	880
5	Ala	Gly	Ala	Gly	λεπ 885		Asn	Ser	Asn	Gly 890		Ser	Val	Ser	Trp 895	
	Glu	ı Pro	Gly			Gln	Val	Pro			Glr	His	Met		Him	
0,	. Leu	ı Ser	Val	900 His		Lvs	Thr	Asn	905 Glu		Ala	CVB	Asn	910 Gln	•	. 11a
			915		•			920	ĺ	•		•	925			
	Val	930	Lys	Pro	Leu	Thr	Lys 935		Tyr	Gln	Gly	940	Gly	Lys	Ser	Leu
5	Thr 945	Phe	Ser	Asp	Thr	Ser 950	Thr	Lys	Thr	Leu	Tyr 955		Val	Glu	Glu	G1u 960
	Glu	Asp	Ala	Gln	Pro 965		Arg	Phe	Ser	Pro 970	Pro	Cly	Ser	Pro	Ser 975	
20	Val	Val	His	Arg			Pro	Ser	Ala	Ala	Thr	Thr	Pro	Pro		
				980			·		985		• • . •			990		*
*	Pro	His	Leu 995	Thr	λla	Glu	,Glu	Thr 100	_	Leu	Phe	Leu	Ala 100		Pro	Ala
25	Leu	Pro 1010		Gly	Leu	Pro	Pro 101		Leu	Gln	Gln	Gln 102	_	Gln	Pro	Pro
	Pro 102	Gln 5	Gln	Lys	Ser	Leu 103		yab	Gln	Leu	Gln 103		Val	Val	Ser	Asn 1040
- T		Ser	Thr	Ala	Ile			Phe	His	Ala			λla	Ċly	Pro	7 : -
10		. •			104					1050		•			105	
	Gly	Pro	Gly	1060	Gly 0	Leu	Arg	Ser	Leu 106		Pro	Pro	Pro	Pro 1070		Pro
9 5	Gln	His	Leu 1075	Gln S	Met	Leu	Pro	Leu 1080		Leu	Ser	Thr	Phe 1085		Glu	Glu
*	Leu	Val 1090	Ser	Pro	Pro	Ala	Asp		Asp	Asp	Asp	Ser 1100		Arg	Phe	Lys
	Leu 110	Leu	Gln	Glu	Tyr			Glu	His	Glu		Glu		λsn	Thr	
10		λар	Glu	Leu	Glu	1110		Glu	Glu	Agn	1119	• •	11 0	31 m	Sar	1120
8			J	,·	112					1130		J	AIG.	n.a	1135	-
	∴ ≀:Leu	Thr	Pro	Asp 1140		Ser	Pro	λla	Lou 1145		Pro	Pro	Ser	Pro 1150		Arg
15	Двр	Ser	Val 1155		Ser	Gly	Ser	Ser 1160		Pro	Ser	Ser			Ser	G1u
	Ser	Val			Thr	Pro	Pro		- 10	Ser	Tyr	Ala	1165 Ser		Ile	Leu
io		1170	•	-" .	٠.	•	1175		*			1180		,*		. –
	1185	yab	ıyr	rys	GIn	Ser 1190		ser	Thr	ren		•	*	10 S		7
•			-													

.19

(3) INFORMATION FOR SEQ ID NO:2:

- (i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 3582 base pairs

 - (B) TYPE: nucleic acid
 - (C) STRANDEDNESS: both (D) TOPOLOGY: linear

(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:

- (A) NAME/KEY: CDS (B) LOCATION: 1..3582

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

	•			•			
	ATGGTCGGGC	TCCTTTIGTT	TTTTTTCCCA	GCGATCTTTT	TOGAGGTGTC	ссттстсссс	60
*	AGAAGCCCCG	GCAGGAAAGT	GTTGCTGGCA	GGAGCGTCGT	CTCAGCGCTC	OGTOGCCAGA	. 120
	ATGGACGGAG	ATGTCATCAT	TCCACCCCTC	TTCTCAGTCC	ATCACCAGCC	TCCGGCCGAG	180
• .	AAAGTGCCCG	AGAGGAAGTG	TGGGGAGATC	AGGGAGCAGT	ATGGCATCCA	GACCGTCGAG	240
	CCCATGTTCC	ACACGTTOGA	TAAGATCAAC	GCGCACCCGG	TCCTCCTGCC	CAACATCACC	300
	CTGGGCAGTG	AGATCCGGGA	CTCCTGCTGG	CACTCTTCCG	TOGCTCTOGA	ACAGAGCATT	360
. ,	GAGTTCATTA	GCGACTCTCT	GATTTCCATT	CGAGATGAGA	AGGATGGGAT	CAACCGGTGT	420
٠.	CTCCCTGACG	GCCAGTCCCT	CCCCCAGGC	AGGACTAAGA	AGCCCATTGC	GGGAGTGATC	480
	OCTCCCGCT	CCAGCTCTGT	AGCCATTCAA	GTGCAGAACC	TGCTCCAGCT	CTTCGACATC	540
	CCCCAGATCG	CTTATTCAGC	CACAAGCATC	GACCTGAGTG	ACAAAACTTT	GTACAAATAC	600
• 00	TTCCTGAGGG	TIGICCCTIC	TGACACTTTG	CAOGCAAGGG	CCATGCTTGA	CATAGTCAAA	660
	CGTTACAATT	GGACCTATGT	CTCTGCAGTC	CACACGGAAG	OGAATTATOG	GGAGAGCGGA	720
,	ATGGACGCTT	TCAAAGAGCT	GCTGCCCAG	GAAGGCCTCT	GTATCGCCCA	TTCTGACAAA	780
	ATCTACAGCA	ACGCTGGGGA	GAAGAGCTTT	GACCGACTCT	TGCGCAAACT	CCGAGAGAGG	840
	CTTCCCAAGG	CTAGAGTGGT	GCTCTGCTTC	TGTGAAGGCA	TGACAGTGCG	AGGACTCCTG	900
	AGCGCCATGC	GCCCCTTGG	сстсстоссс	GAGTTCTCAC	TCATTGGAAG	TGATOGATOG	960
•	GCAGACAGAG	ATGAAGTCAT	TGAAGGTTAT	GAGGTGGAAG	CCAACGGGGG	AATCACGATA	1020
`. •	AAGCTGCAGT	CTCCAGAGGT	CACCTCATTT	GATGATTATT	TCCTGAAACT	GAGGCTGGAC	1080
	ACTAACACGA	CGAATCCCTG	GTTCCCTGAG	TTCTGGCAAC	ATCOGTTCCA	GTGCCGCCTT	1140
· •	CCAGGACACC	TTCTGGAAAA	TCCCAACTTT	AAACGAATCT	GCACAGGCAA	TGALAGCTTA	1200
	GAAGAAAACT	ATGTCCAGGA	CAGTAAGATG	GCCTTTCTCA	TCAATGCCAT	CTATGCCATG	1260
	GCACATGGGC	TGCAGAACAT	GCACCATGCC	стстсссстс	GCCACGTGGG	CCTCTGCGAT	1320
	GCCATGAAGC	CCATCGACGG	CAGCAAGCTG	CTGGACTTCC	TCATCAAGTC	CTCATTCATT	1380
.00	GGAGTATCTG	GAGAGGAGGT	CTCCTTTCAT	GAGAAAGGAG	ACGCTCCTGG	AAGGTATGAT	1440
	ATCATGAATC	TGCAGTACAC	TGAAGCTAAT	CGCTATGACT	ATGTGCACGT	TOGAACCTOG	1500

		CATGAAGGAG	TGCTGAACAT	TGATGATTAC	AAAATCCAGA	TGAACAAGAG	TOGACTOCTC	1560
		COGTCTGTGT	GCAGTGAGCC	TTGCTTAAAG	GGCCAGATTA	AGGTTATACG	GAAAGGAGAA	1620
5	-	GTGAGCTGCT	GCTGGATTTG	CACGGCCTGC	AAAGAGAATG	AATATGTGCA	AGATGAGTTC	1680
•		ACCTGCAAAG	CTTGTGACTT	COGATOCTCC	CCCAATGCAG	ATCTAACAGG	CTGTGAGCCC	1740
		ATTCCTGTGC	GCTATCTTGA	GTGGAGCAAC	ATCGAACCCA	TTATAGCCAT	CCCCTTTTCA	1800
		TGCCTGGGAA	TCCTTGTTAC	СТТСТТТСТС	ACCCTAATCT	TTGTACTGTA	CCCCCACACA	1860
10		CCAGTGGTCA	AATCCTCCAG	TCGGGAGCTC	TGCTACATCA	TCCTAGCTGG	CATCTTCCTT	1920
	. *	OCTTATCTCT	GCCCATTCAC	TCTCATTGCC	AAACCTACTA	CCACCTCCTG	CTACCTCCAG	1980
		CCCCTCTTCC	TTGGCCTCTC	CTCTGCGATG	TOCTACTCTC	CTTTAGTGAC	TAAAACCAAT	2040
15		CGTATTGCAC	GCATCCTGGC	TOGCAGCAAG	AAGAAGATCT	GCACCCCGAA	GCCCAGGTTC	2100
	•	ATGAGTGCCT	GGGCTCAGGT	GATCATTCCC	TCAATTCTGA	TTAGTGTGCA	ACTAACCCTG	2160
	. •	GTGGTAACCC	TGATCATCAT	GGAACCCCCT	ATGCCCATTC	TGTCCTACCC	AAGTATCAAG	2220
20		GAAGTCTACC	TTATCTGCAA	TACCAGCAAC	CTGGGTGTGG	TOCCCCCTTT	GGGCTACAAT	2280
		OGACTCCTCA	TCATGAGCTG	TACCTACTAT	GCCTTCAAGA	CCCGCAACGT	GCCCGCCAAC	2340
		TTCAACGAGG	CCAAATATAT	CGCGTTCACC	ATGTACACCA	CCTGTATCAT	CTGGCTAGCT	2400
25		TTTGTGCCCA	TTTACTTTCC	GAGCAACTAC	AAGATCATCA	CAACTTGCTT	TGCAGTGAGT	2460
		CTCAGTGTAA	CAGTOGCTCT	OCCUTCCATC	TTCACTCCCA	AGATGTACAT	CATTATTGCC	2520
•		AAGCCTGAGA	CCAATCTCCC	CAGTGCCTTC	ACCACCTCTG	ATGTTGTCCG	CATGCATGTT	2580
		GOCGATOGCA	ACCTGCCCTG	CCCCTCCAAC	ACTITICATION	ACATCTTCCG	AAGAAAGAAG	2640
30		GCAGGGGCAG	CCAATCCCAA	TTCTAATGGC	AAGTCTGTGT	CATOGTCTGA	ACCAGGTGGA	2700
		OGACACGTCC	-CCAAGGGACA	GCATATGTGG	CACCGCCTCT	CTGTGCACGT	GAAGACCAAT	2760
*		GAGACGCCCT	GCAACCAAAC	ACCCCTCATC	AAACCCCTCA	CTAAAAGTTA	CCAAGGCTCT	2820
35	0	GGCAAGAGCC	TGACCTTTTC	AGATACCAGC	ACCAAGACCC	TTTACAACGT	AGAGGAGGAG	2880
		GAGGATGCCC	AGCCGATTCG	CTTTAGCCCG	CCTCGTAGCC	CTTCCATOGT	GCTGCYCYCC	2940
*		CGCGTGCCAA	GCGCGGCGAC	CACTCCGCCT	CTGCCGCCCC	ACCTGACCGC	AGAGGAGACC	3000
40		CCCCTCTTCC	TOGCCGAACC	AGCCCTCCCC	AAGGCTTGC	CCCCTCCTCT	CCAGCAGCAG	3060
•		CAGCAACCCC	CTCCACAGCA	GAAATCGCTG	ATGGACCAGC	TCCAGGGAGT	GGTCAGCAAC	3120
		TTCAGTACCG	CGATCCCGGA	TTTTCACGCG	GTGCTGGCAG	eccccoooce	TCCCOOGAAC	3180
- 45	•	COCCTCCCCT	CCCTGTACCC	GCCCCCCCCA	CCTCCGCAGC	ACCTGCAGAT	CCTCCCCCTC	3240
		CAGCTGAGCA	CCTTTGGGGA	GGAGCTGGTC	TCCCCGCCCG	CGGACGACGA	CGACGACAGC	3300
	*	GAGAGGTTTA	AGCTCCTCCA	GGAGTACGTG	TATGAGCACG	AGCGGGAAGG	GAACACCGAA	3360
• • •	. &	GAAGACGAAC	TGGAAGAGGA	GGAGGAGGAC	CTGCAGGCGG	CCAGCAAACT	GACCCCCGAT	3420
.50		GATTCGCCTG	CGCTGACGCC	TCCGTCGCCT	TTCCGCGACT	COCTOCCCTC	GGGCAGCTCG	3480

		CTG	CCAG	CT CC	CCAGT	GTC (CGAGT	CGGTG	CIC	TGCA	ccc (CTCC	CXXC	GT A	TCCT	'ACGC	С	354	0
	,	TCT	STCAT	TC TG	CGGGA	CTA C	CAAGC.	AAAGC	TCT	TCCA	ccc	TG	:			••		358	2
5								٠		•					•				٠ _
		(4)	INFO	RMATI	ON FO	r seç	Q ID	NO:3:											:
.10	8		(i)	(A) (B) (C)	ENCE LENG TYPE STRA	TH: 6 : nuc NDEDN	60 ba: cleic: NESS:	se pa acid both	irs		*						٠.		
				(D)	TOPO	LOGY :	: lin	ear	•			·	•					٠.	
	•	•	(ii)	MOLE	CULE	TYPE	cDN	A		•									
			(ix)	PEAT	URE:	ς	•	2 10				,					•		
15			,,		NAME	/KEY:	: CDS												
				(B)	LOCA	TION:	: 1	50	•		,								
*			(xi)	SEQU	ENCE	DESC	RIPTI	ON: S	eq I	D NO	:3:				,				,
		3 mai		00 ma		~~~	~~ `	Mac 1	^aaa	a mormo		· •	~~~	T C C	~~~	mccc	~		
20		ATG	TCGG	GC IV	CTTTT	GTT /I	1717171	ICCCA	GCG.	AICT.	1-1-1-	1 GGA	3016 .	ic c	CITC	ICCC	-		6
				,	-					•	•	•	•			•	•		٠.٠.
		(5)	INFO	RMATI	ON FO	R SEC	Q ID I	NO:4:	•										
			(i)	SEQU	ENCE	CHARA	ACTER:	ISTIC	S:	·									
25		-			LENG			-								;			
23			•	• •	TYPE							•		•					
٠	•				STRA					٠.									
	*1		(ii)	MOLE	CULE	TYPE:	cDN							·		-			
30			(ix)	FEAT	URE:								· ·						٠.
·.	(· (,	(A)	NAME			50	•			• •	•	•					9
			(xi)	SEQU	ENCE	DESCR	RIPTIO	ON: SI	EQ II	D NO:	:4:-		•				:		
35		CCAC	GACA	CC TT	CTGGA	AAA T	rccca)	CTTT	AAA	ÇG AA 1	rci (CAC	VGGC2	u n	GAAN	GCTT/	\ , '		60
						•							•						
. *		(6)	INFO	RMATI	ON FO	R SEC	ID I	ю:5:,			• :			0	•		•	-	2 _, +
40 ´			(i)		ENCE LENG											•			
`~		,	*	(B)	TYPE	: nuc	cleic	acid					•						
-	•	.*		(D)	TOPO	LOGY:	line	ear	·		9	•					•		. ,
45			(ii)	MOLE	CULE	TYPE:	cDN/	.	-					•	٠.	٠.			
• 1		÷	(ix)	FEAT				•								•			•
•		*			NAME LOCA			50	٠.				• •0		•	· · .	•	0	
50 50			(xi)	SEQU	ENCE	DESCR	RIPTIO	N: SI	EQ II	D N O:	5:	÷				1.		٠.	
		AACG	TATO	CT AC	GCCTC	TGT C	ATTC	rccc	GAC.	TAC A	AGC 1	LAAGO	TCT	C C1	ACCC:	IGTA	Χ	60	

SEQUENCE LISTING

		y ± 7	GLITE	-					•					-			•	
		•	(i)	(B)	ICAN STR	REET: TY:	Li Indi	lly ianap	Corp	orat							•.	
·)				(E)	STA COU	NTRY	?: L		ed St	ates	of	Amer	ica		,	٠	•	•
•		`	(ii)		E OF							trop	oic G	luta	umate	Rec	epto	r
			(iii)	NUM	BER C	F SE	EQUE	CES:	: 5				٠,					
5			(iv)	(A)	RESPO	RESS	SEE:	C. N	4. Hu		 1		*	1 ***				
	•			(C)	CIT	Y: V	Windl	lesha										. '
,				(E)	STA COU	NTR	e: Ur	nited	l Kir	ngdon	n	• .		• .				• • •
	•		· (v)		PUTER							6						
				(B)	MEE COM	1PUT	ER: N	dack i	intos	sh								
		•			OPE SOF							17.0				:		
5	•			(0)												•		
		(2)	INFO	RMAT.	ION. F	FOR S	SEQ :	ID NO	0:1:	•			•					٠.
	•		(i)	(A)	JENCE) LEN TYP	NGTH PE: 6	: 119 amino	4 ar	nino id		is							
	•			(D)	TOF	POLO	GY:	linea	ar .	•		•	٠	•				
			-(ii)	MOLI	ECULE	E TY	PE: I	prote	ein							•	•.	
	•		(xi)	SEQ	JENCE	E DES	SCRI	PTIO	V: SI	EQ II	D NO	:1:			*			
5	. •		Met 1	Val	G1y	Leu	Leu 5	Leu	Phe	Phe	Phe	Pro 10	Ala	Ile	Phe	Leu	Glu 15	Va1
			Ser	Leu	Leu	Pro 20	Arg	Ser	Pro	Gly	Arg 25	Lys	Val	Leu	Leu	Ala 30	Gly	Ala
)	•		Ser	Ser	Gln 35	Arg	Ser	Val	Ala	Arg 40	Met	Asp	Gly	Asp	Val 45	Ile	Ile	Gly ·
· .			Ala	Leu 50	Phe	Ser	Val	His	His 55	Gln	Pro	Pro	Ala :	Glu 60	Lys	Val	Pro	Glu
5	•		Arg 65	ГЛа	Суз	Gly	Glu	11e 70	Arg	Glu	Gln	Tyr	Gly 75	Ile	Gln	Arg	Val	G1u 80
		1 . 1	Ala	Met	Phe	His	Thr 85	Leu	Asp	Lys	Ile	Asn 90	Ala	Asp	Pro	Val	Leu 95	Leu
_			Pro	Asn	Ile	Thr 100	Leu	Gly	Ser	Glu	Ile 105	Arg	Asp	Ser	Сув	Trp 110	His	Ser
9			Ser	Val	Ala 115	Leu	Glu	Gln	Ser	Ile 120	Glu	Phe	Ile	Arg	Asp 125	Ser	.Leu	Ile

	. GAAGACGAAC TOGAAGAGGA GGAGGAGGAC CTGCAGGCGG CCAGCAAACT GACCCCGGAT	3420
	GATTCGCCTG CGCTGACGCC TCCGTCGCCT TTCCGCGACT CGGTGGCCTC GGGCAGCTCG	3480
	GTGCCCAGCT CCCCAGTGTC CGAGTCGGTG CTCTGCACCC CTCCCAACGT ATCCTACGCC	3540
5	TCTGTCATTC TGCGGGACTA CAAGCAAAGC TCTTCCACCC TG	3582
		3 18
	(4) INFORMATION FOR SEQ ID NO:3:	
		,
10	(i) SEQUENCE CHARACTERISTICS: (A) LENGTH: 60 base pairs	
	(B) TYPE: nucleic acid	
	(C) STRANDEDNESS: both (D) TOPOLOGY: linear	•
•		
15	(ii) MOLECULE TYPE: CDNA	
	(ix) FEATURE:	
•	(A) NAME/KEY: CDS (B) LOCATION: 160	
	(b) LOCATION: 11.60	•
20	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:3:	0.0
	ATGGTCGGGC TCCTTTTGTT TTTTTTCCCA GCGATCTTTT TGGAGGTGTC CCTTCTCCCC	60
*	(5) INFORMATION FOR SEQ ID NO:4:	
0.5	(i) SEQUENCE CHARACTERISTICS:	
25	(A) LENGTH: 60 base pairs	
	(B) TYPE: nucleic acid (C) STRANDEDNESS: both	*
	(D) TOPOLOGY: linear	
	(ii) MOLECULE TYPE: cDNA	
30		. :
	(ix) FEATURE: (A) NAME/KEY: CDS	
	(B) LOCATION: 160	
	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:	e e
35	CCAGGACACC TTCTGGAAAA TCCCAACTTT AAACGAATCT GCACAGGCAA TGAAAGCTTA	
	CCAGGACACC TICIGGARAA TCCCAACTIT AAACGAATCT GCACAGGCAA TGAAAGCTTA	
	(6) INFORMATION FOR SEQ ID NO:5:	w d
,	(6) INFORMATION FOR SEQ ID NO. 3.	
40	(i) SEQUENCE CHARACTERISTICS: (A) LENGTH: 60 base pairs	. 9
	(B) TYPE: nucleic acid	
	(C) STRANDEDNESS: both (D) TOPOLOGY: linear	
		×
45	(ii) MOLECULE TYPE: cDNA	
-	(ix) FEATURE:	
	(A) NAME/KEY: CDS (B) LOCATION: 160	
•		
50	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:	
50	AACGTATCCT ACGCCTCTGT CATTCTGCGG GACTACAAGC AAAGCTCTTC CACCCTGTAA	60

		Ser	Ile 130	Arg	Asp	Clu		Asp 135	Gly	Ile	Asn	Arg	Cys 140	Leu	Pro	Asp	Gly .	
5		Gln 145	Ser	Leu	Pro		Gly 150	Arg	Thr	Lys	Lys	Pro 155	Ile	Ala	Gly	Val.	Ile 160	
J	-8-	Gly	Pro	Gly	Ser	Ser 165	Ser	Val.	Ala ·	Ile	Gln 170	Val	Gln	Asn	Leu	Leu 175	Gln	٠
٠.		Leu	Phe	Asp	Ile 180	Pro	Gln	Ile	Ala	Tyr 185	Ser	Ala	Thr	Ser	11e 190	Asp	Leu	
10		Ser	Asp	Lys 195	Thr	Leu	Tyr	Lys	Tyr 200	Phe	Ĺeu	Arg	Val	Val 205	Pro	Ser	Asp	
		Thir	Leu 210	Gln	Ala	Arg	Ala	Met 215	Leu	Asp	Ile	Val	Lys 220	Arg	Tyr	Asn	Trp	•
15		Thr 225	Tyr	Val	Ser	Ala	Val. 230	His	Thr	Glu	Gly	Asn 235	Tyr	Gly	Glu	Ser	Gly 240	
		Met	Asp	Ala	Phe	Lys 245	Glu	Leu	Ala	Ala	Gln 250	Glu	Gly	Leu	Суз	Ile 255	Ala	
20		His	Ser	Asp	Lys 260	Ile	туr	Ser	Asn	Ala 265	Gly	Glu	Ļys	Ser	Phe 270	Asp	Arg	
		Leu	Leu	Arg 275	Lys	Leu	Arg	Glu	Arg 280	Leu	Pro	ГЛа	Ala	Arg 285	Val	Val	Val	
25		Cys	Phe 290	СЛа	Glu	Gly	Met	Thr 295	Val	Arg	Gly	Leu	Leu 300	Ser	Ala	Met	Àrg	
	•	Arg 305	Leu	Gly	Val	Val	Gly 310	Glu	Phe	Ser	Leu	Ile 315	Gly	Ser	Asp	GĴĄ	Trp 320	
30		Ala	Asp	. Arg	Asp	Glu 325	Val	Ile	Glu	Gly	Tyr 330	Glu	Val	Glu	Ala	Asn 335	Gly	
		Gly	Ile	Thr	Ile 340	Lys	Leu	Gln	Ser	Pro	Glu	Val	Arg	Ser	Phe 3'50	Asp	Asp	
		Tyr	Phe	Leu 355	Lys	Leu	Arg	Leu	Asp 360	Thr	Asn	Thr	Arg	Asn 365	Pro	Trp	Phe	
35		Pro	Glu 370		Ťrp	Gln	His	Arg 375		Gln	Суз	Arg	Leu 380	Pro	Gly	His	Leu	
		Leu 385		Asn	Pro) Asn	Phe 390	Lys	Arg	Ile	Cys	Thr 395	Gly	Asn	Glu	Ser	Leu 400	
40		Glu	Glu	Asn	Tyr	Val 405	Gln	Asp	Ser	Lys	Met 410	Gly	Phe	Val	Ile	Asn 415	Ala	
										•							· · · · ·	
		· Ile	Tyr	Ala	Met 420	: Ala	His	Gly	Leu	Glr 425	Asn S	Met	His	His	Ala 430	Leu	Cys	
45		:			420 Val				Asp	425	•		•		Asp		Ser	
45		Pro	Gly	His 435	420 Val	Gly	⁄. Leu	Cys	Asp 440	425 Ala	Met	Lys	Pro	Ile 445	Asp	Gly		-
45		Pro	Gly Lev 450	His 435 Leu	420 Val	Gly	/ Leu	Cys Ile 455	Asp 440 Lys	Ala Ala Ser	Met Ser	Lys Phe	Pro	Ile 445	Asp Val	Gly Ser	Ser	

•												,					
	11	e Met	Asn		Gln 485	Tyr	Thr	Glu		Asn 490	Arg	Tyr	Asp	Tyr	Val 495	His	٠
5 · .	Va.	l Gly	Thr	Trp 500		Glu	Gly	Val	Leu 505	Asn	Ile	Asp	qeA	Tyr 510	ГЛЗ	Ile	,
	Gl	n Met	Asn 515	Lys	Ser	Gly	Val	Val 520		Ser	Val	Cys	Ser 525	Glu	Pro	СХё	
10		u Lys 530	Gly	Gln	Ile	Ly.s	Val 535		Arg	Lys	Gly	Glu 540	Val	Ser	Суз	СУз	
		p Ile	Cys	Thr	Ala	Cys 550	Lys	Glu	Asn	Glu	Tyr 555	Val	Gln	Asp	Glu	Phe 560	
•		r Cys	Lys	Ala	Cys 565	,	Leu	Gly	Trp	Trp 570	Pro	Asn	Ala	Aab	Leu 575	Thr	
15	Gl	у Суз	Glu	Pro 580		Pro	Val	Arg	Tyr 585		Glu	Trp	Ser	Asn 590		Glu	
	Pr	o Ile			Ile	Alá	Phe			Leu	Gly	Ile	Leu 605		Thr	Leu	
20	Ph	e Val		Leu	İle	Phe		600 Leu	Tyr	Arg	Asp	Thr		Val	Val	Lys	
	Se	610 r Ser		Arg	Glu	Leu	615 Cys	Tyr	Ile	 Ile	Leu	620 Ala	Gly	Ile	Phe	Leu	•
25	62 G1	5 y Tyr	Val	Cvs	Pro	630 Phe	Thr	Leu	Ile		635 Lys	Pro	Thr	Thr	Thr	640 Ser	
		s Tyr			645	•	. 8		•	650		•	•	1 .	655		٠
*-	•	_		660	•				665					670 . ,			•
-30		r Ala	675				*	680					685		·		
	Se	r Lys 690	7 -	Lys	Ile	CÀa	Thr 695	Arg	Lys	Pro	Arg	Phe 700	Met	Ser	Ala	Trp	
35	A1 70	a Gln 5	Val	Ile	Tle	Ala 710	Ser	Ile	Leu	Ile	Ser 715	Val	Gln	Leu	Thr	Leu 720	
	· Va	l Val	Thr	Leu	Ile 725		Met	Glu	Pro	Pro 730	Met	Pro	Ile	Leú	Ser 735	Tyr	
40	Pr	o Ser	Ile	Lys 740	Glu	Val	Tyr	Leu	11e 745	Суз	Asn	Thr	Ser	Asn 750	Leu	Gly	
*	Va.	l Val	Ala 755		Leu	Gly	Tyr	Asn 760	Gly	Leu	Leu	Ile	Met 765	Ser	Cys	Thŗ	
45	Tγ	r Tyr 770		Phe	Lys	Thr	Arg 775	Asn	Val	Pro	Ala	Asn 780	Phe	Asn	Glu	Ala	
s	Ly 78	s Tyr 5	Ile	Ala	Phe	Thr 790	Met	Tyr	Thr	Thr	Cys 795	Ile	Ile	Trp	Leu	Ala 800	
	Ph	e Val	Pro	Ile	Týr 805	Phe	Gly	Ser	Asn	Tyr 810		Ile	Ile	Thr	Thr 815	CÀa	
50	Ph	e Ala	Val	Ser 820	Leu	Ser	Val	Thr	Val 825	Ala	Leu	Gly	СХа	Met 830	Phe	Thr	
•		•								,						• ,	

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		,		Pro		Met 83 5		Ile	Ile	Ile	Ala 840 -	Lys	Pro	Glu	Arg	Asn 845	Val	Arg	Ser
5	•				Phe 850	Thr	Thr	Ser	Asp	Val= 855	Val	Arg	Met	His	Val 860	Gly	Asp	ĠĮĄ	ГЛЗ
				Leu 865		Cys	Arg	Ser	Asn 870	Thr	Phe	Leu	Asn	Ilė 875	Phe	Arg	Arg	Ļys	880 FAa
10	-,	•		Ala	Glý	Ala	Gly	Asn 885	Ala	Asn	Ser	Asn	Gly 890	ГЛЗ	Ser	Val	Ser	Trp 895	Ser
		•		Glu	Pro	Gly	Gly 900	Gly	Gln	Val	Pro	Lys 905	Gly	Gln	His	Met	Trp 910	His	Arg
15				Leu	Ser	Val 915	His	Val	Lys	Thr	Asn 920	Glu	Thr	Ala	Cys	Asn 925	Gln	Thr	Ala
, ,					11e 930	Lys	Pro	Leu	Thr	Lys 935		Tyr	Gln.	Gly	Ser 940	Gly	Lys	Ser	Leu
				Thr 945	Phe	Ser	Asp	Thr	Ser 950	Thr	Lys	Thr	Leu	Tyr 955	Asn	Val	Glu	Glu	Glu .
20				Glu	Asp	Ala	Gln	Pro 965	Ile	Arg	Phe	Ser	Pro 970	Pro	Gly	Ser	Pro	Ser 975	Met
				Val	Val	His	Arg. 980	Arg	Väl	Pro	Ser	Ala 985	Ala	Thr	Thr	Pro	Pro 990	. Leu	Pro
25			. •.*	Pro	His	Leu 995	Thr	Ala	Glu	Glu	Thr 1000	Pro	Leu	Phe	Leu	Ala 100	Glu 5	Pro	Ala
	,		,	Leu	Pro 101		Gly	Leu	Pro	Pro 101	Pro	Leu	Gln	Gln	Gln 102	Gln 0	Gln	Pro	Pro
<i>30</i>				Pro 1025		Gln	Lys	Ser	Leu 103	Met 0	Asp	Gln	Leu	Gln 103	Gly 5	Val	Val	Ser	Asn 1040
	• • • • • • • • • • • • • • • • • • • •	•		Phe	Ser	Thr	Ala	11e 104		Asp	Phe	His	Ala 105	Val 0	Leu	Ala	Gly	Pro 105	Gly 5
35		•		Gly	Pro	Cly	Asn 106		Leu	Arg	Ser	Leu 106	Tyr 5	Pro	Pro	Pro	Pro 107	Pro 0	Pro
• •				Gln	His	Leu 107		Met	Leu	Pro	Leu 108	Gln 0	Leu	Ser	Thr	Phe 108	Gly 5	Glu	Glu
40				Leu	Val 109	Ser 0	Pro	Pro	Ala	Asp 109	Asp 5	Asp	Asp	Asp	Ser 110	·Glu 0	Arg	Phe	Lys
·			The .	Leu 110		Gln	Glu	Tyr	Val		Glu	His	Glu	Arg 111	Glu 5	Gly	Asn	Thr	Glu 1120
45				Glu	Asp	Glu	Leu	Glu 112		Glu	Glu	Glu	Asp 113	Leu 0	Gln	Ala	Ala	Ser 113	Lys 5
•	· · · · · · · · · · · · · · · · ·		•	Leu	Thr	Pro	Asp 114		Ser	Pro	Ala	Leu 114	Thr 5	Pro	Pro	Ser	Pro 115	Phe 0	Arg
				Asp	Ser	.Val		Ser	Gly	Ser	Ser 116	Val 0	Pro	Ser	Ser	Pro 116	Val	Ser	Glu
50 			•	Ser	Val 117		Cys	Thr	Pro	Pro 117	neA 'S	Val	Ser	Tyr	'Ala 118	. Ser 10	Val	: Ile	Leu
																			•

Arg Asp Tyr Lys Gln Ser Ser Ser Thr Leu 1185

(3) INFORMATION FOR SEQ ID NO:2:

- (i) SEQUENCE CHARACTERISTICS:
 - (A) LENGTH: 3582 base pairs
 - (B) TYPE: nucleic acid
 - (C) STRANDEDNESS: both
- (D) TOPOLOGY: linear

(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:

- (A) NAME/KEY: CDS
- (B) LOCATION: 1..3582

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(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

ATGGTCGGCC	TCCTTTTGTT	TTTTTTCCCA	GCGATCTTTT	TGGAGGTGTC	CCTTCTCCCC	60
AGAAGCCCCG	GCAGGAAAGT	GTTGCTGGCA	GGAGCGTCGT	ÇTCAGCGCTC	GGTGGCCAGA	120
ATGGACGGAG	ATGTCATCAT	TGGAGCCCTC	TTCTCAGTCC	ATCACCAGCC	TCCGGCCGAG	180
AAAGTGCCCG	AGAGGAAGTG	TGGGGAGATC	AGGGAGCAGT	ATGGCATCCA	GAGGGTGGAG	240
GÉCATGTTCC	ACACGTTGGA	TAAGATCAAC	GCGGACCCGG	TCCTCCTGCC	CAACATCACC	300
CTGGGCAGTG	AGATCCGGGA	CTCCTGCTGG	CACTCTTCCG	TGGCTCTGGA	ACAGAGCATT	360
GAGTTCATTA	GGGACTCTCT	GATTTCCATT	CGAGATGAGA	AGGATGGGAT	CAACCGGTGT	420
CTGCCTGACG	GCCAGTCCCT	CCCCCAGGC	AGGACTAAGA	AGCCCATTGC	GGGAGTGATC	480
GGTCCCGGCT	CCAGCTCTGT	AGCCATTCAA	GTGCAGAACC	TGCTCCAGCT	CTTCGACATC	540
CCCCAGATCG	CTTATTCAGC	CACAAGCATC	GACCTGAGTG	ACAAAACTTT	GTACAAATAC	600
TTCCTGAGGG	TTGTCCCTTC	TGACACTITG	CAGGCAAGGG	CCATGCTTGA	CATAGTCAAA	660
CGTTACAATT	GGACCTATGT	CTCTGCAGTC	CACACGGAAG	GGAATTATGG	GGAGAGCGGA	720
ATGGACGCTT	TCAAAGAGCT	GGCTGCCCAG	GAAGGCCTCT	GTATCGCCCA	TTCTGACAAA	780
ATCTACAGCA	ACGCTGGGGA	GAAGAGCTTT	GACCGACTCT	TCCCCAAACT	CCGAGAGAGG	840
.CTTCCCAAGG	CTAGAGTGGT	GGTCTGCTTC	TGTGAAGGCA	TGACAGTGCG	AGGACTCCTG	, 900
AGCGCCATGC	GGCGCCTTGG	CGTCGTGGGC	GAGTTCTCAC	TCATTGGAAG	TGATGGATGG	960
GCAGACAGAG	ATGAAGTCAT	TGAAGGTTAT	GAGGTGGAAG	CCAACGGGGG	AATCACGATA	1020
AAGCTGCAGT	CTCCAGAGGT	CAGGTCATTT	GATGATTATT	TCCTGAAACT	GAGGCTGGAC	1080
ACTAACACGA	GGAATCCCTG	GTTCCCTGAG	TTCTGGCAAC	ATCGGTTCCA	GTGCCGCCTT	1140
CCAGGACACC	TTCTGGAAAA	TCCCAACTTT	AAACGAATCT	GCACAGGCAA	TGAAAGCTTA	1200
GAAGAAAACT	ATGTCCAGGA	CAGTAAGATG	GGGTTTGTCA	TCAATGCCAT	CTATGCCATG	1260
GCACATGGGC	TGCAGAACAT	GCACCATGCC	стстсссстс	GCCACGTGGG	CCTCTGCGAT	1320
GCCATGAAGC	CCATCGACGG	CAGCAAGCTG	CTGGACTTCC	TCATCAAGTC	CTCATTCATT	1380

	GGAGTATCTG	GAGAGGAGGT	GTGGTTTGAT	GAGAAAGGAG	ACGCTCCTGG	AAGGTATGAT	1440 -
	ATCATGAATC	TGCAGTACAC	TGAAGCTAAT	CGCTATGACT	ATGTGCACGT	TGGAACCTGG	1500
5	CATGAAGGAG	TGCTGAACAT	TGATGATTAC	AAAATCCAGA	TGAACAAGAG	TGGAGTGGTG	1560
7	CGGTCTGTGT	GCAGTGAGCC	TTGCTTAAAG	GGCCAGATTA	AGGTTATACG	GAAAGGAGAA	1620
	GTGAGCTGCT	GCTGGATTTG	CACGGCCTGC	AAAGAGAATG	AATATGTGCA	AGATGAGTTC	1680
10	ACCTGCAAAG	CTTGTGACTT	GGGATGGTGG	CCCAATGCAG	ATCTAACAGG	CTGTGAGCCC	1740
	ATTCCTGTGC	GCTATCTTGA	GTGGAGCAAC	ATCGAACCCA	TTATAGCCAT	CGCCTTTTCA	1800
	TGCCTGGGAA	TCCTTGTTAC	CTTGTTTGTC	ACCCTAATCT	TTGTACTGTA	CCGGGACACA	1860
• 00	CCAGTGGTCA	AATCCTCCAG	TCGGGAGCTC	TGCTACATCA	TCCTAGCTGG	CATCTTCCTT	1920
15	GGTTATGTGT	GCCCATTCAC	TCTCATTGCC	AAACCTACTA	CCACCTCCTG	CTACCTCCAG	1980
	сесстстте	TTGGCCTCTC	CTCTGCGATG	TGCTACTCTG	CTTTAGTGAC	TAAAACCAAT	- 2040
	CGTATTGCAC	GCATCCTGGC	TGGCAGCAAG	AAGAAGATCT	GCACCCGGAA	GCCCAGGTTC	2100
20	ATGAGTGCCT	GGGCTCAGGT	GATCATTGCC	TCAATTCTGA	TTAGTGTGCA	ACTAACCCTG	2160
	GTGGTAACCC	TGATCATCAT	GGAACCCCCT	ATGCCCATTC	TGTCCTACCC	AAGTATCAAG	2220
	GAAGTCTACC	TTATCTGCAA	TACCAGCAAC	CTCCCTCTCC	TGGCCCCTTT	GGGCTACAAT	2280
25	GGACTCCTCA	TCATGAGCTG	TACCTACTAT	GCCTTCAAGA	CCCGCAACGT	GCCCGCCAAC	2340
	TTCAACGAGG	ССАААТАТАТ	CGCGTTCACC	ATGTACACCA	CCTGTATCAT	CTGGCTAGCT	2400
	TTTGTGCCCA	TTTACTTTGG	GAGCAACTAC	AAGATCATCA	CAACTTGCTT	TGCAGTGAGT	2460
30	CTCAGTGTAA	CAGTGGCTCT	GGGGTGCATG	TTCACTCCCA	AGATGTACAT	CATTATTGCC	2520
	AAGCCTGAGA	GGAATGTCCG	CAGTGCCTTC	ACCACCTCTG	ATGTTGTCCG	CATGCATGTT	2580
	GGCGATGGCA	AGCTGCCCTG	CCGCTCCAAC	ACTTTCCTCA	ACATCTTCCG	AAGAAAGAAG	2640
as :	GCAGGGGCAG	GGAATGCCAA	TTCTAATGGC	AAGTCTGTGT	CATGGTCTGA	ACCAGGTGGA	2700
35	GGACAGGTGC	CCAAGGGACA	GCATATGTGG	CACCGCCTCT	CTCTCCACCT	GAAGACCAAT	2760
	.GAGACGGCCT	GCAACCAAAC	AGCCGTCATC	AAACCCCTCA	CTAAAAGTTA	CCAAGGCTCT	2820
	GGCAAGAGCC	TGACCTTTTC	AGATACCAGO	ACCAAGACCC	TTTACAACGT	AGAGGAGGAG	2880
40	GAGGATGCCC	AGCCGATTCG	CTTTAGCCCG	CCTGGTAGCC	CTTCCATGGT	GGTGCACAGG	2940
*	CGCGTGCCAA	GCGCGGCGAC	CACTCCGCCT	CTGCCGCCCC	ACCTGACCGC	AGAGGAGACC	3000
*	CCCCTCTTCC	TGGCCGAACC	AGCCCTCCCC	AAGGGCTTGC	CCCCTCCTCT	CCAGCAGCAG	3060
45	CAGCAACCCC	CTCCACAGCA	GAAATCGCTG	ATGGACCAGC	TCCAGGGAGT	GGTCAGCAAC	3120
·	TTCAGTACCG	CGATCCCGGA	TTTTCACGCG	GTGCTGGCAG	GCCCGGGGG	TCCCGGGAAC	3180
	GGGCTGCGGT	CCCTGTACCC	GCCCCCGCCA	CCTCCGCAGC	ACCTGCAGAT	C. GCTGCCGCTG	3240
50	CAGCTGAGCA	CCTTTGGGGA	GGAGCTGGTC	TCCCCGCCCG	CGGACGACGA	CGACGACAGC	3300
	GAGAGGTTTA	AGCTCCTCCA	GGAGTACGTC	TATGAGCACG	AGCGGGAAGG	GAACACCGAA	3360
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55 Claims

1. A human metabotropic glutamate receptor protein which comprises the amino acid sequence SEQ ID NO:1 or functional equivalents thereof.

- 2. The human metabotropic glutamate receptor protein of Claim 1 which is SEQ ID NO:1.
- A nucleic acid compound which comprises a nucleic acid sequence that encodes all or part of the protein of Claim 1.
- 4. The nucleic acid compound of Claim 3 which is SEQ ID NO:2.
- 5. A recombinant DNA vector which comprises the nucleic acid compound of Claim 3 or Claim 4.
- 10 6. A host cell transfected with the recombinant DNA vector of Claim 5.
 - 7. The transfected host cell of Claim 6 which is E. coli/pRS117.
- 8. A method for determining whether a substance interacts with or affects the protein of SEQ ID NO:1, said method comprising contacting a functional compound of SEQ ID NO:1 with said substance, monitoring interaction by physically detectable means, and identifying those substances which effect a chosen response.
 - 9. A method for constructing a host cell capable of expressing the protein of Claim 1, said method comprising transfecting a host cell with a recombinant DNA vector that comprises the nucleic acid compound of Claim 3, and culturing said host cell under conditions suitable for the expression of said nucleic acid compound.
 - 10. A method for expressing a nucleic acid compound which encodes SEQ ID NO:1 or a functional equivalant thereof in a host cell transfected with said nucleic acid compound, said method comprising culturing said transfected host cell of Claim 6 under conditions suitable for expression of said nucleic acid compound.

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Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.5)
D, X	NATURE. vol. 349, 28 February 1991, LONDON GB pages 760 - 765 Masu M; Tanabe Y; Tsuchida K; Shigemoto R; Nakanishi S; 'Sequence and expression of a metabotropic glutamate receptor.' * the whole document *	1-10	C12N15/12 C07K13/00 C12N1/21 G01N33/68, //(C12N1/21, C12R1:19)
D,X	SCIENCE vol. 252, 31 May 1991, LANCASTER, PA pages 1318 - 1321	1-10	
	Houamed KM; Kuijper JL; Gilbert TL; Haldeman BA; O'Hara PJ; Mulvihill ER; Almers W; Hagen FS; 'Cloning, expression, and gene structure of a G protein-coupled glutamate receptor from rat brain.' * the whole document *		*
>,χ	WO-A-9 210 583 (ZYMOGENETICS INC, US) 25 June 1992	1-10	-
	* the whole document *	,	TECHNICAL FIELDS SEARCHED (Int. Cl.5)
			C12N C07K G01N
-		je S	
, .	The present search report has been drawn up for all claims	2	
-	Place of search THE HAGUE O3 AUGUST 1993	<u> </u>	NAUCHE S.A.
X: par Y: par doc A: tec O: no	CATEGORY OF CITED DOCUMENTS T: theory or princip E: earlier patent do after the filing d ticularly relevant if combined with another Unment of the same category A: mers ber of the s ticularly relevant descriptions L: document cited if L: document cited if the disclosure d: mers ber of the s document	le underlying the cument, but publi ate in the application or other reasons	invention isbed on, or

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